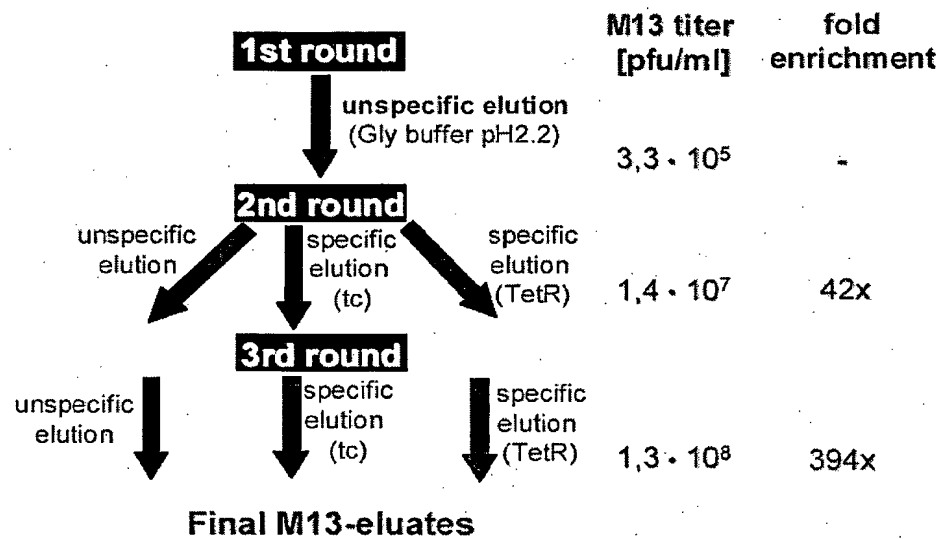


**Figure 1: Experimental procedure for the *in vitro* selection.**

**Figure 2: Example for *in vitro* selected sequences.**

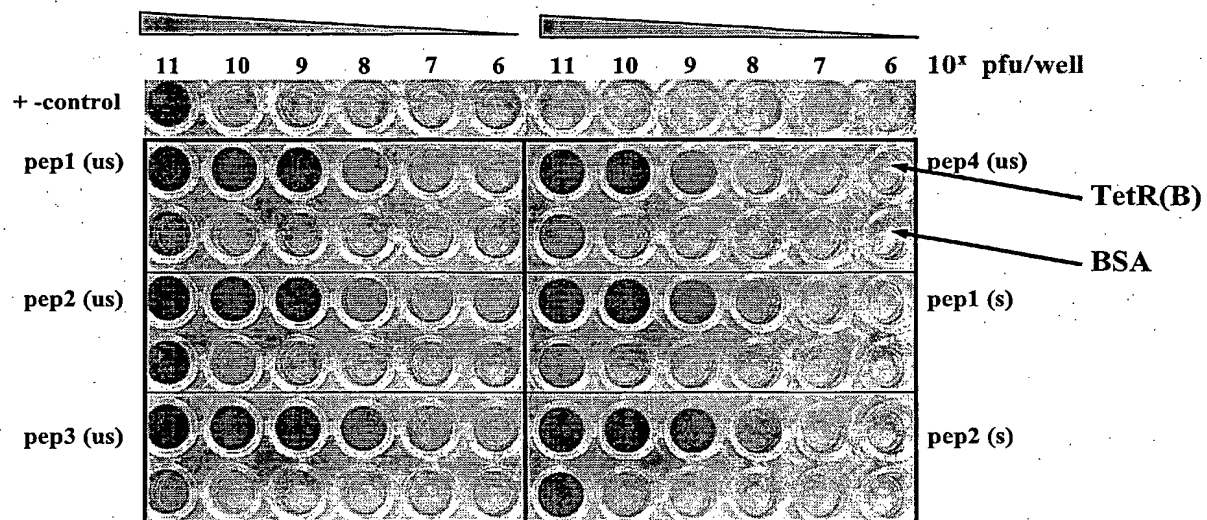
- Unspecific elution (Gly buffer, pH 2.2)

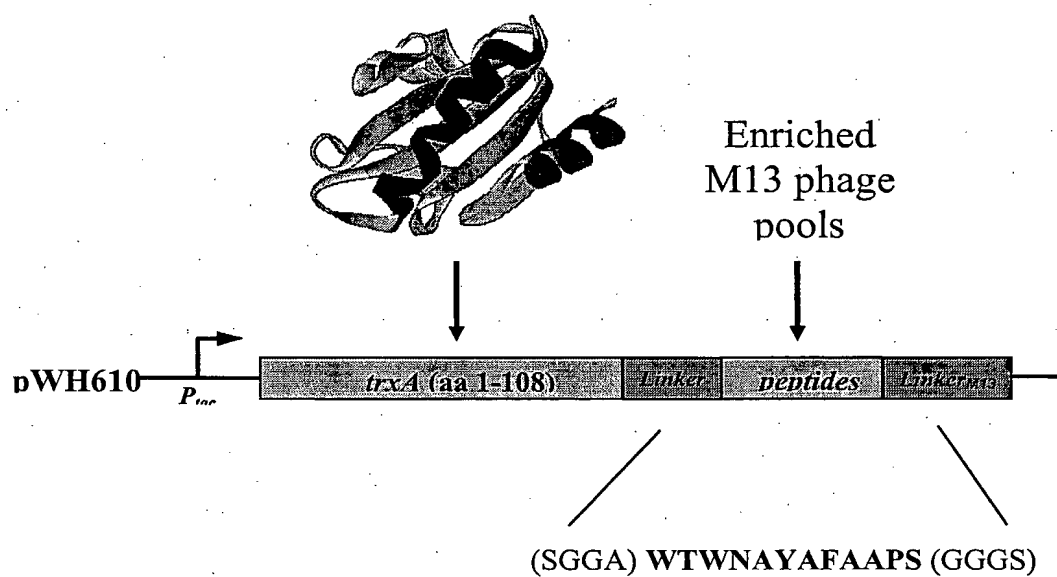
pep1	Trp	-	His	-	Gly	-	Ala	-	Ile	-	Leu	-	Gly	-	Ser	-	Ala	-	Arg	-	Ala	-	Gln
pep2	Leu	-	Pro	-	Ser	-	Tyr	-	Met	-	Leu	-	His	-	Leu	-	Trp	-	Ser	-	Pro	-	His
pep3	Ala	-	His	-	Tyr	-	Ser	-	Leu	-	Tyr	-	Trp	-	Pro	-	Met	-	Ala	-	Thr	-	Phe
pep4	Tyr	-	His	-	Asn	-	Leu	-	Tyr	-	Gly	-	Leu	-	Pro	-	Leu	-	Gly	-	Pro	-	Trp
pep5	Trp	-	His	-	Gln	-	Thr	-	Tyr	-	Thr	-	Ser	-	Ser	-	Leu	-	Trp	-	Glu	-	Ser

- Specific elution (TetR, 4 $\mu$ M)

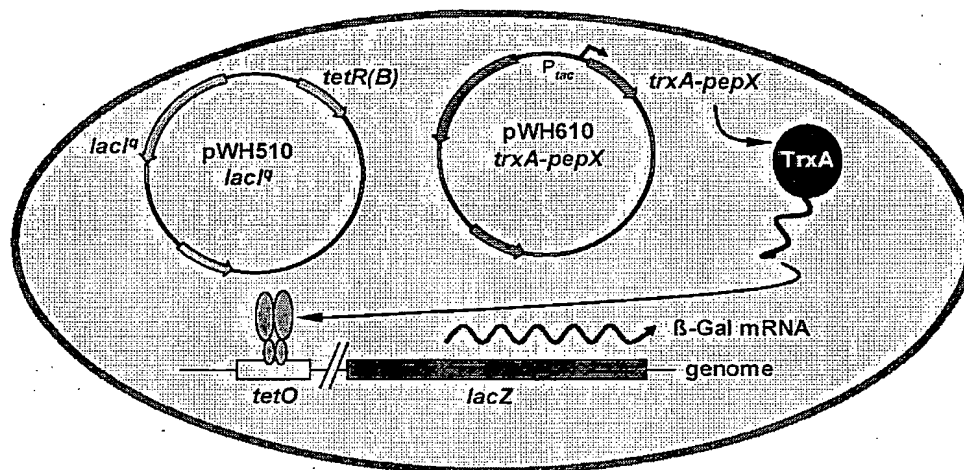
pep1	Trp	-	Thr	-	Trp	-	Asn	-	Ala	-	Tyr	-	Ala	-	Phe	-	Ala	-	Ala	-	Pro	-	Ser
pep2	Trp	-	His	-	Ser	-	Ser	-	Phe	-	Asn	-	Met	-	Phe	-	Ala	-	Tyr	-	Pro	-	Met
pep3	Trp	-	His	-	Leu	-	Pro	-	Leu	-	Ser	-	Trp	-	Thr	-	Thr	-	Arg	-	Leu	-	Pro
pep4	Trp	-	His	-	Thr	-	Pro	-	Ile	-	Ser	-	Leu	-	Leu	-	Lys	-	Gln	-	Val	-	Arg
pep5	Trp	-	His	-	Trp	-	Thr	-	Phe	-	Ser	-	Ser	-	Pro	-	Leu	-	Met	-	Gln	-	Thr

**Figure 3: Characterisation of TetR-phage binding by ELISA.**



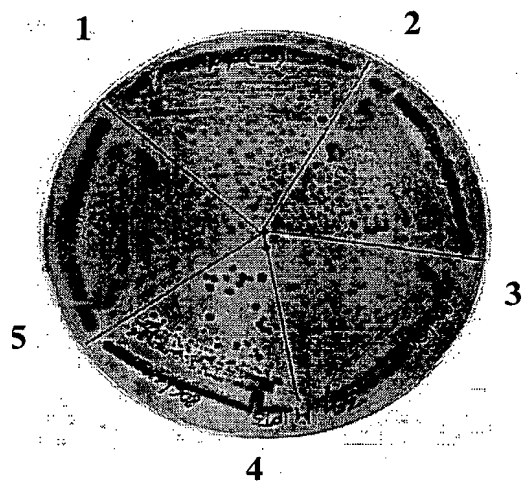
**Figure 4: Design of the peptide expressing construct.**

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**Figure 5: Setup of the *in vivo* screening system.**

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Figure 6: McConkey plate.



box	Plasmid I* encoding	Plasmid II** encoding	$\beta$ -Gal activity
1	TetR(B)	TrxA-pepBs1	+
2	TetR(B)	TrxA-pepBs1	+
3	TetR(B)	-	-
4	-	-	+(100%)
5	TetR(B)	TrxA	-

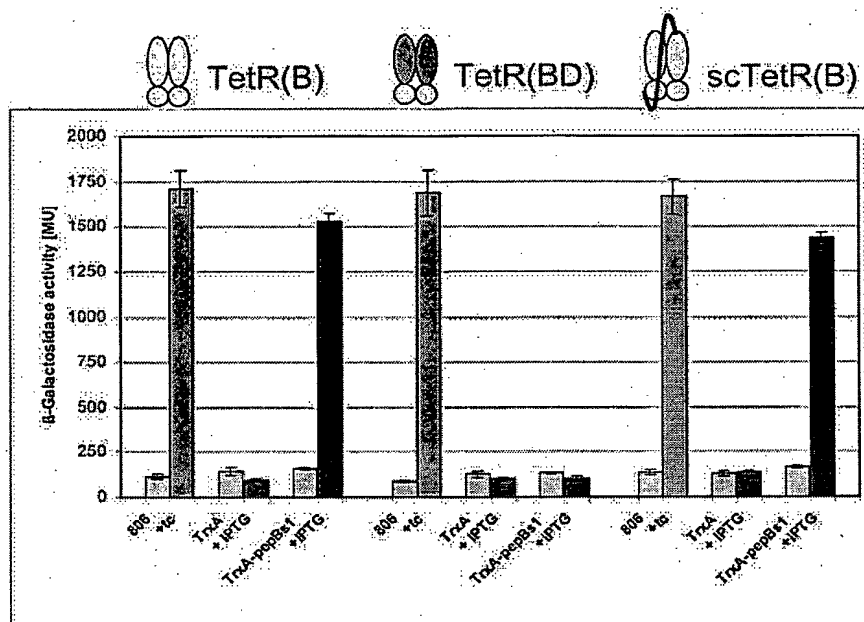
\* pWH510*lacZ* for TetR(B), pWH1200 (Altschmied et al., 1988)

\*\* pWH610 for TrxA/TrxA-pepBs1, pWH806 (Wissmann et al., 1991)

"+" = induced (yellow colonies)

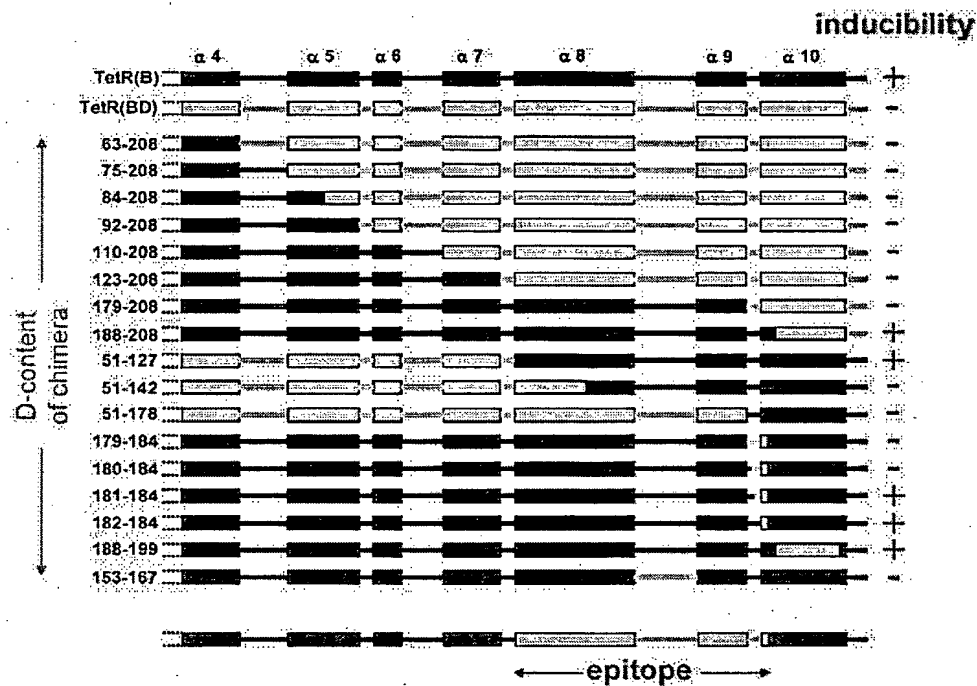
"- " = uninduced (colorless colonies)

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**Figure 7: LacZ assay for the TetR-inducing fusion protein TrxA-pepBs1.**

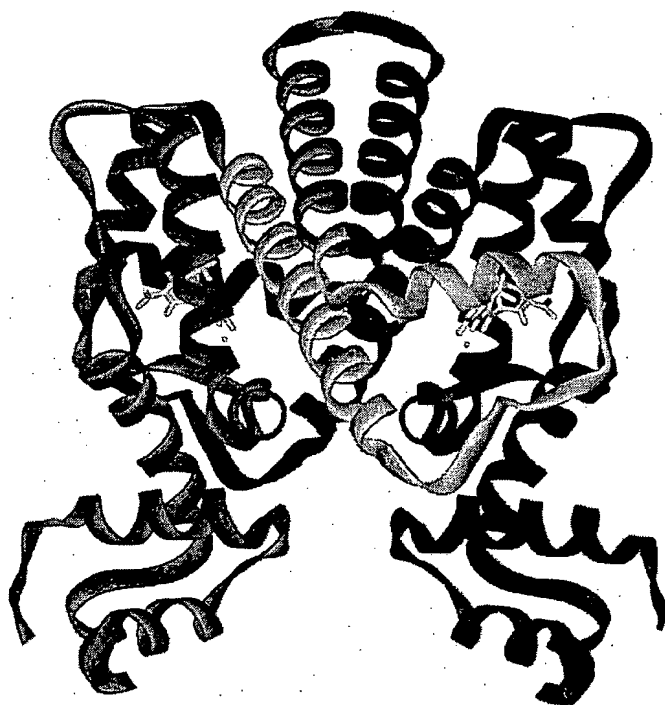
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**Figure 8: Identification of the region of interaction between TetR and TrxA-pepBs1 by *in vivo* epitope mapping.**

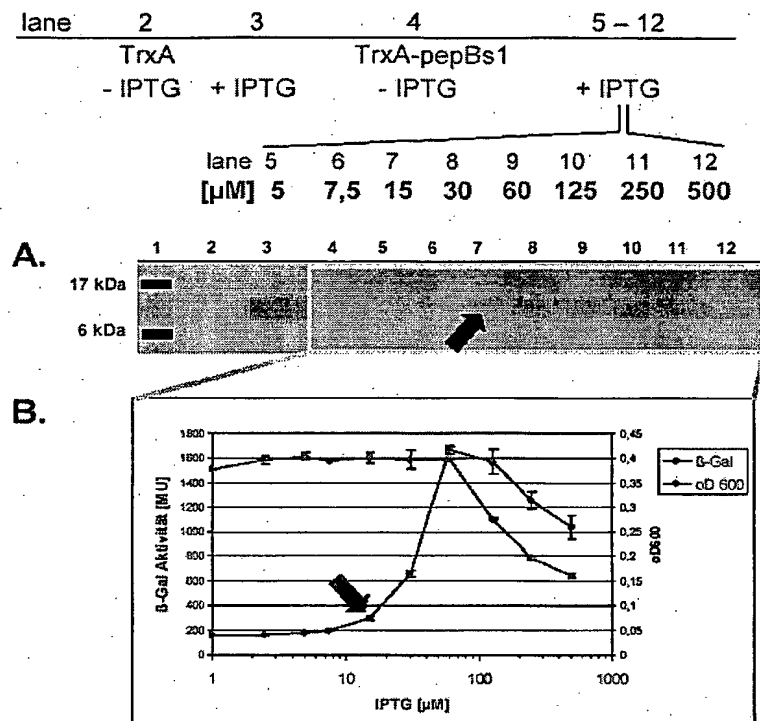




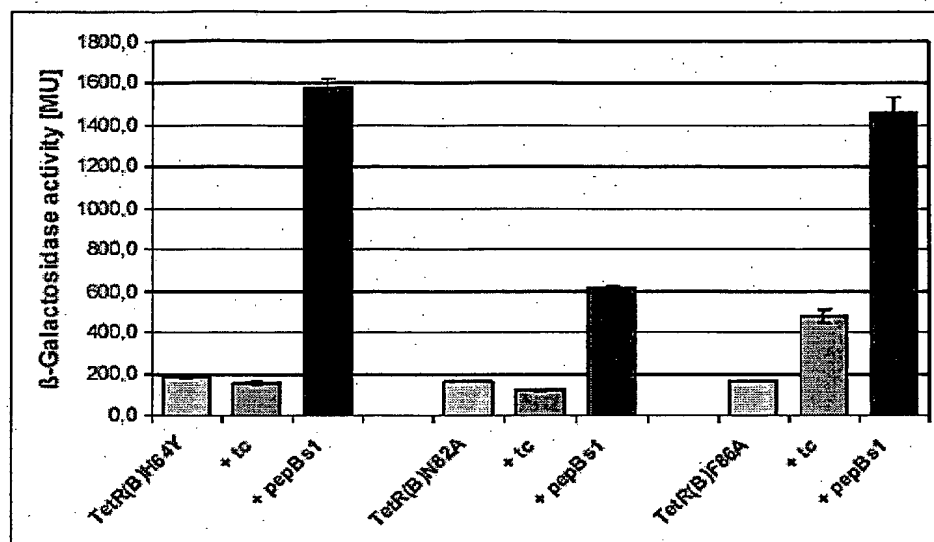
**Figure 9: Structure of TetR.**



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**Figure 10: Expression of the peptide correlates with induction of TetR.**

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**Figure 11: *In vivo* characterisation of non-inducible TetR mutants.**

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**Figure 12:** Position of the amino acids H64, N82 and F86 relative to tetracycline and the interaction epitope.

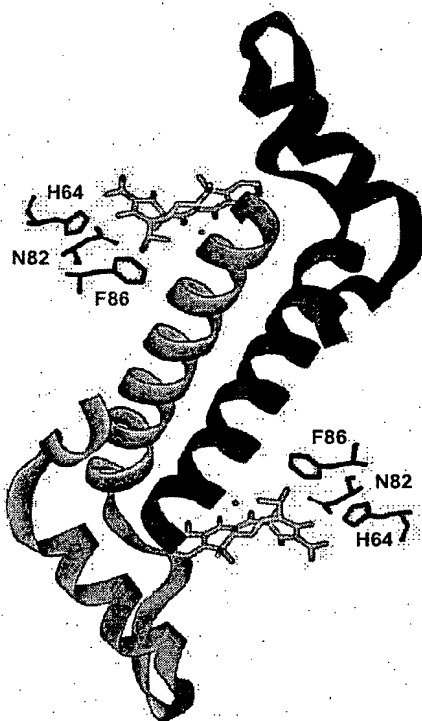
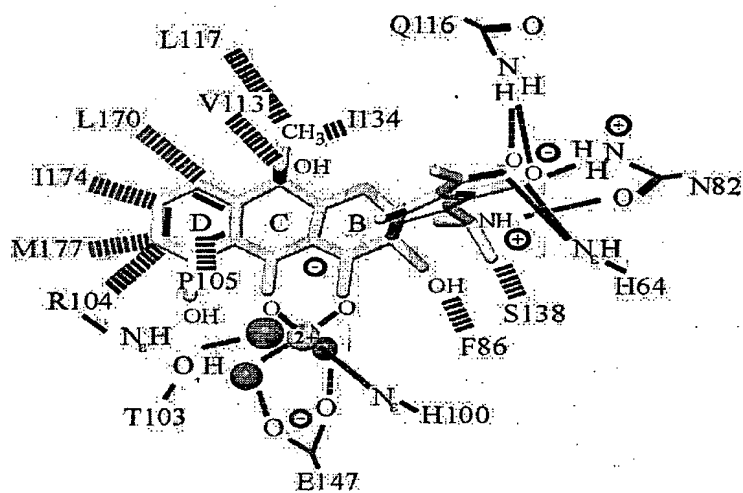
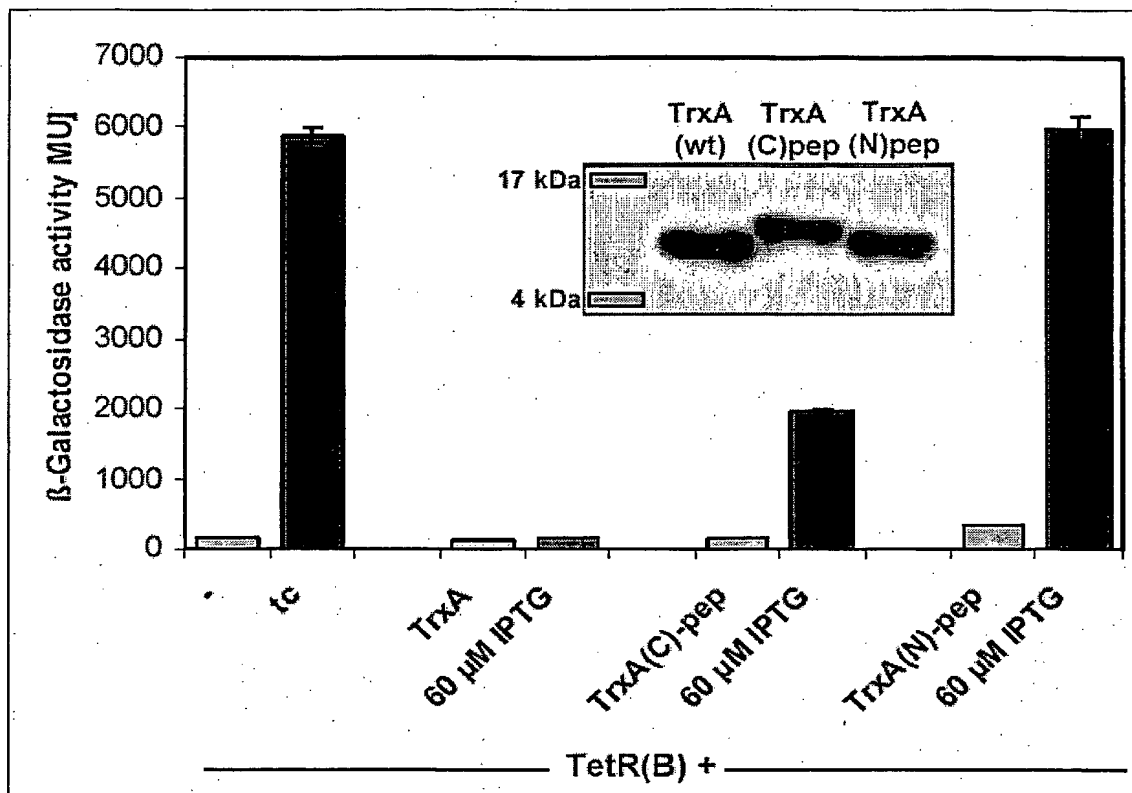


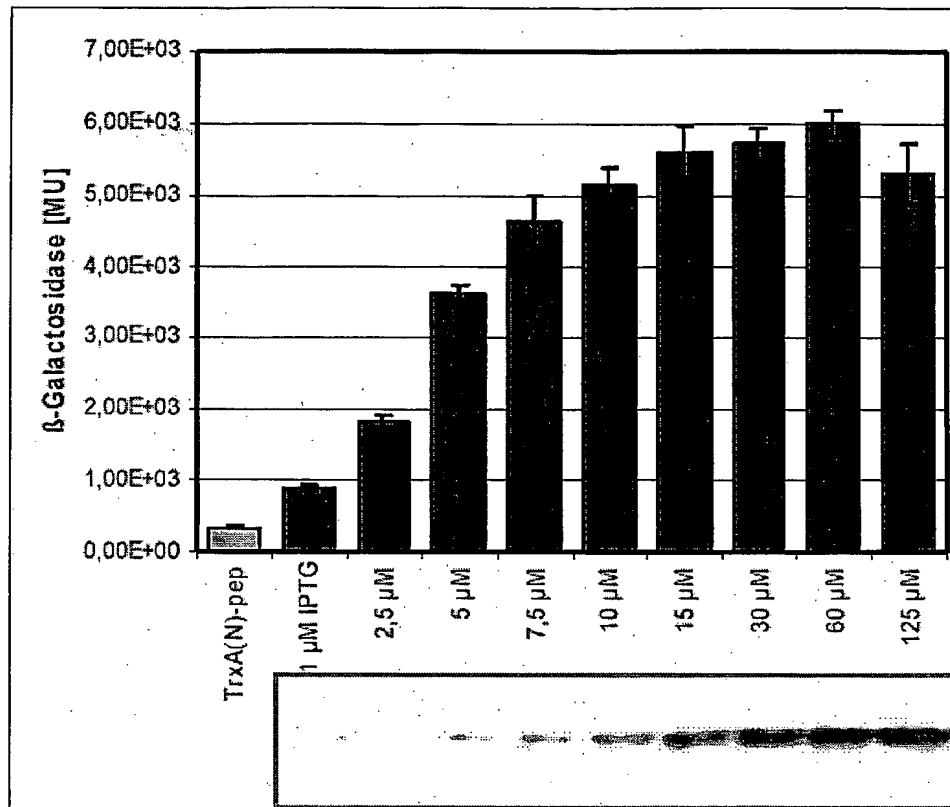
Figure 13: Amino acids contacting tc.

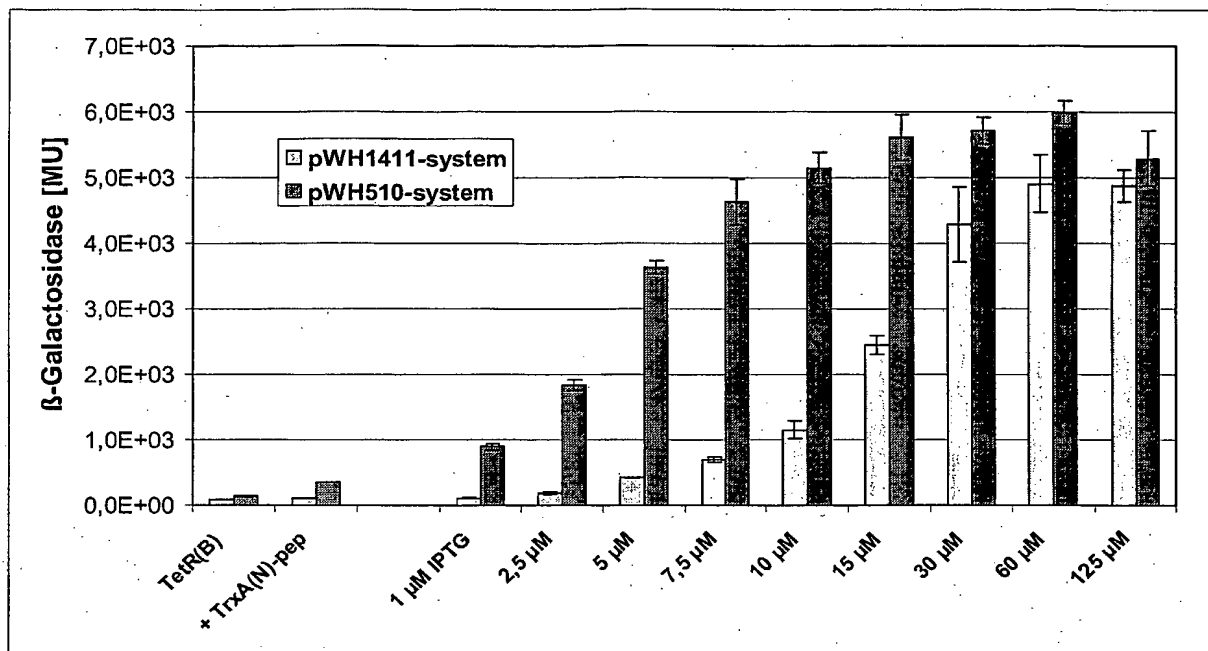


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**Figure 14: *In vivo* characterisation of TetR inducibility by TrxA fusion proteins.**

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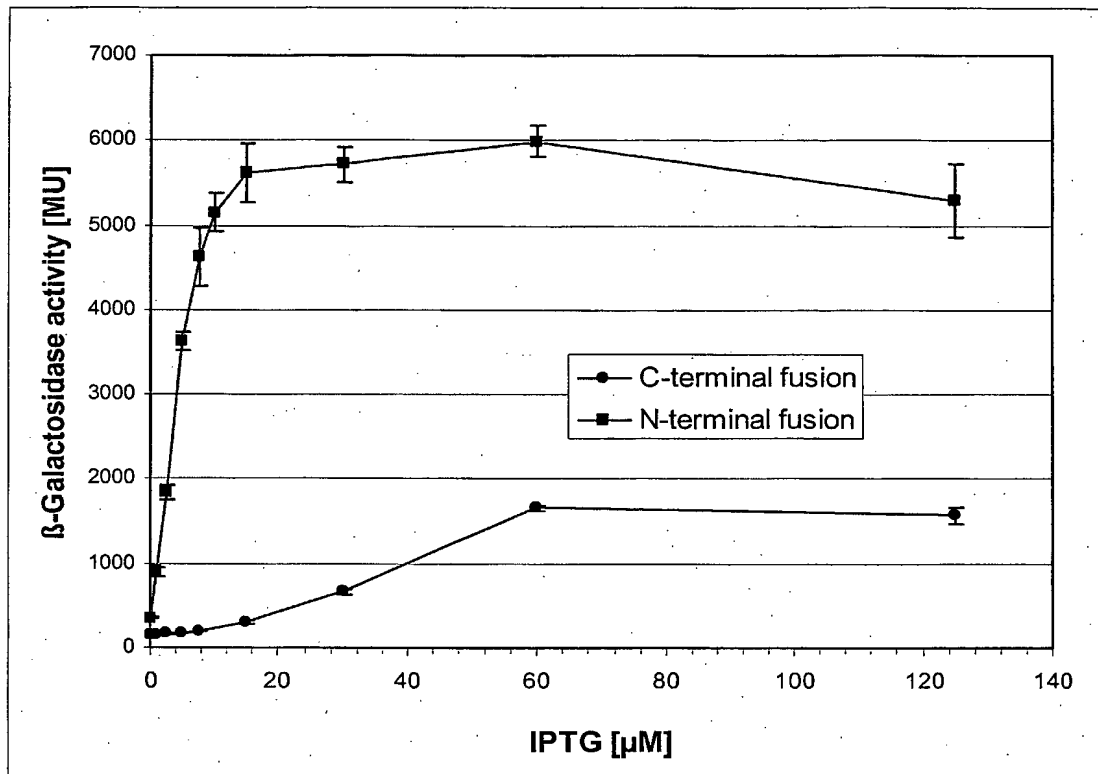
**Figure 15: Correlation between the protein level and induction of TetR(B).**

**Figure 16: Comparison of a low and high TetR-expressing system.**



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Figure 17: Comparison of TetR(B) induction by C- and N-terminal TrxA-peptide fusions.



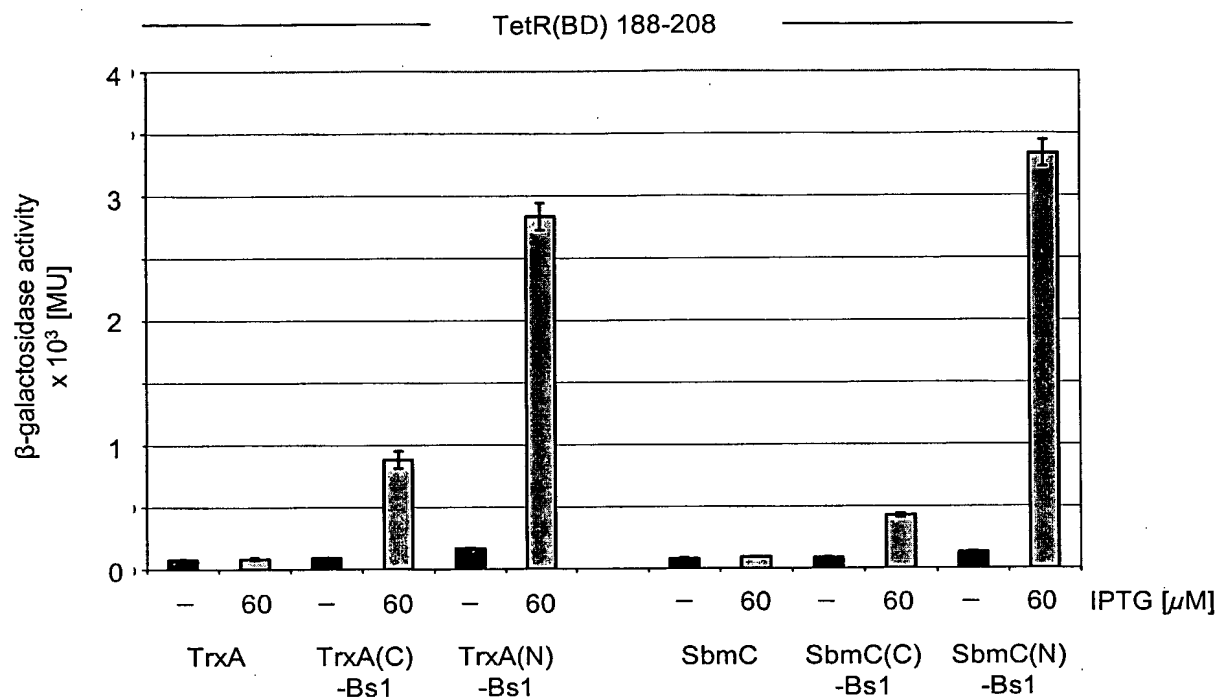
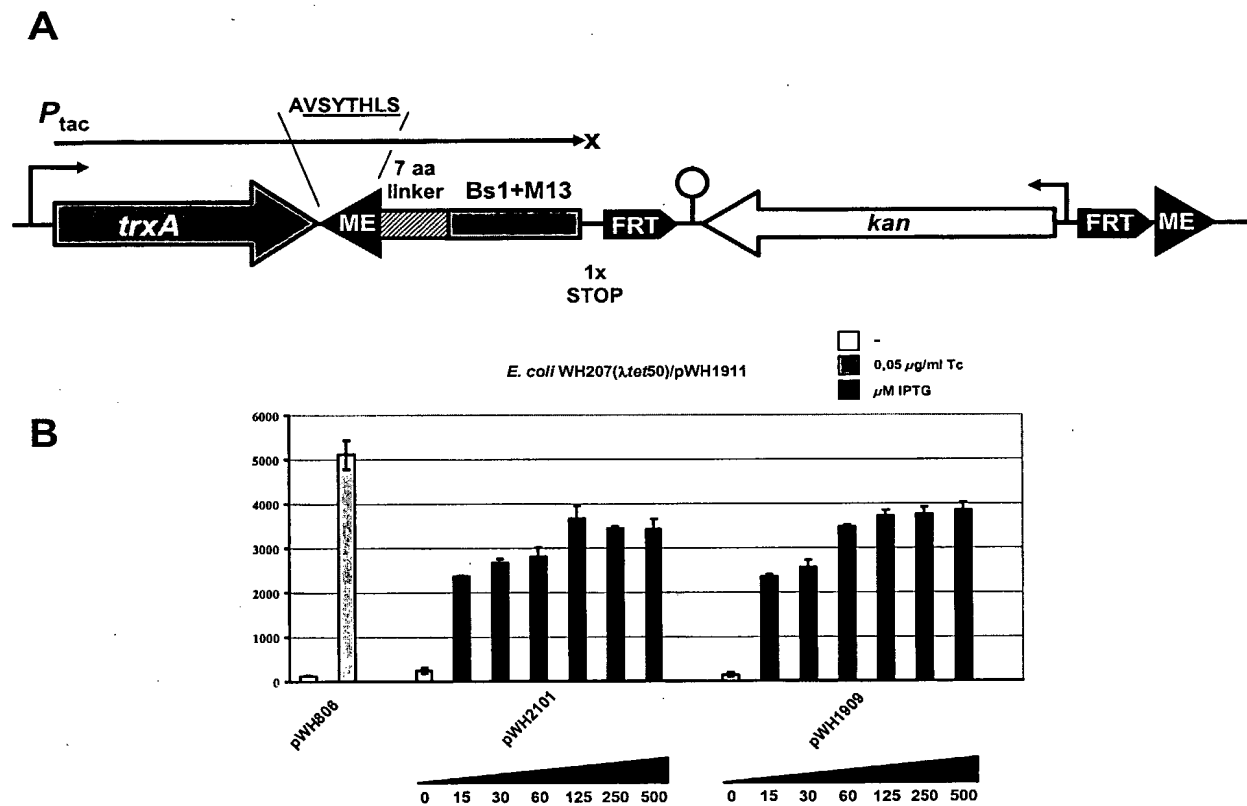
**Figure 18: LacZ assay for the TetR-inducing fusion protein SbmC-pepBs1.**

Figure 19: An in-frame fusion of an insertion element (IE<sup>FKS</sup>) encoding the peptide Bs1 to TrxA leads to a protein that induces TetR(B).



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Figure 20: An in-frame fusion of the insertion element  $IE^{FSK}$  to the *atpD* ORF at its endogenous location in the *E. coli* genome leads to a protein that induces TetR(B).

